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SEROLOGICAL REACTIONS: THE RHEUMATOID FACTOR IN RHEUMATOID ARTHRITIS*

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THE general subject of serological reactions in rheumatoid arthritis could include topics broader in scope than the "rheumatoid factor". The antinuclear serum factors and the biological false positive Wassermann antibodies, though more often features of systemic lupus erythematosus, seem significantly associated with rheumatoid arthritis. The incidence of antinuclear (L.E.) factors in patients with rheumatoid arthritis varies with the serological means of detection and is complicated by uncertainties in the arbitrary clinical and pathologic separation of connective tissue syndromes, but there is no doubt that sera of some patients with "classical" rheumatoid arthritis exhibit antinuclear reactivity.[1, 2]

One serum protein is observed with such frequency in rheumatoid arthritis as to justify the term "rheumatoid factor". This discussion will be limited to the various serological reactions which are attributed to the "rheumatoid factor".

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TABLE I.—THE CHARACTER OF SIX SYSTEMS UTILIZED FOR DETECTION OF THE RHEUMATOID FACTOR

System	*Particulate Carrier*	*"Reactant"*	*References*
Sensitized sheep cell agglutination	sheep erythrocyte	rabbit gamma globulin (hemolysin)	4, 5
Sensitized Rh cell agglutination	Rh-positive human erythrocyte	human gamma globulin (incomplete isoantibody)	8, 9
FII tanned cell agglutination	sheep erythrocyte	human gamma globulin (Cohn FII)	6
FII Bentonite flocculation	Bentonite particle	human gamma globulin (Cohn FII)	10
FII latex fixation	latex particle	human gamma globulin (Cohn FII)	7
FII precipitin reaction	none	human gamma globulin (Cohn FII)	12

Only a brief historical review can be presented here. The streptococcal agglutination test, in 1932, first suggested a role for the beta hemolytic streptococcus in the pathogenesis of rheumatoid arthritis.[3] Subsequent years have not offered support for such a thesis. Bacterial agglutination (streptococci and others) by rheumatoid sera is mediated by the rheumatoid factor. In 1948, Rose, Ragan and associates described the sensitized sheep cell test, in which a high proportion of rheumatoid sera agglutinated sheep erythrocytes coated with rabbit anti-sheep cell hemolysin (in essence, the reagent used in complement fixation procedures).[4] Waller's description of this phenomenon in the Scandinavian literature in 1940 was lost in the confusion of World War II.[5] Currently applied tests for the rheumatoid factor have derived from studies of sensitized sheep cell agglutinations. Heller noted that human Cohn Fraction II (gamma globulin) inhibited sensitized sheep agglutination by rheumatoid sera and subsequently demonstrated that tanned sheep erythrocytes coated with Cohn FII were agglutinated by rheumatoid sera and that this reaction paralleled sensitized sheep cell agglutination.[6] Agglutination of latex particles coated with Cohn FII by rheumatoid sera was described by Singer and Plotz.[7] The widespread application of this procedure is attributed to its relative simplicity and ease of per-

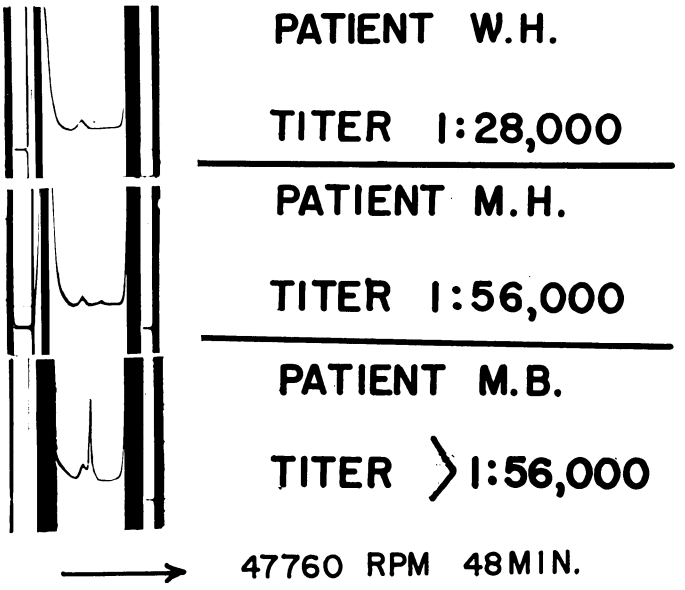

Fig. 1. Analytical centrifuge patterns of three rheumatoid sera with varying titers in FII tanned cell agglutination test. At the time of exposure, serum W. H. shows a single component with an S constant of 19 (not different from normal sera). Sera with higher titers (M.H. and M.B.) demonstrate, in addition to a 19S component, components sedimenting more rapidly.

formance. Many variations of these basic reactions have been proposed but, in essence, they can all be considered as having a common basis. In all instances, a serum protein—the "rheumatoid factor"—reacts with a part of, or a component of, gamma globulin. The gamma globulin factor has been termed the "reactant". The character of several systems is summarized in Table I. In the sensitized sheep cell test, the "reactant" is rabbit immune gamma globulin (hemolysin). Rh-positive human erythrocytes coated with incomplete Rh antibody are agglutinated by the RF.[8, 9] In this instance, the reactant is human immune gamma globulin. The most widely applied tests such as the latex fixation or bentonite flocculation tests[10] utilize pooled normal human Cohn FII as the reactant gamma globulin.

To consider first the rheumatoid factor, this was known for several years to be associated with the gamma globulin fraction of serologically positive rheumatoid sera. Franklin, Kunkel and associates were first to characterize the rheumatoid factor as residing in the high molecular weight fraction of gamma globulin (macroglobulin).[11] Figure 1 illus-

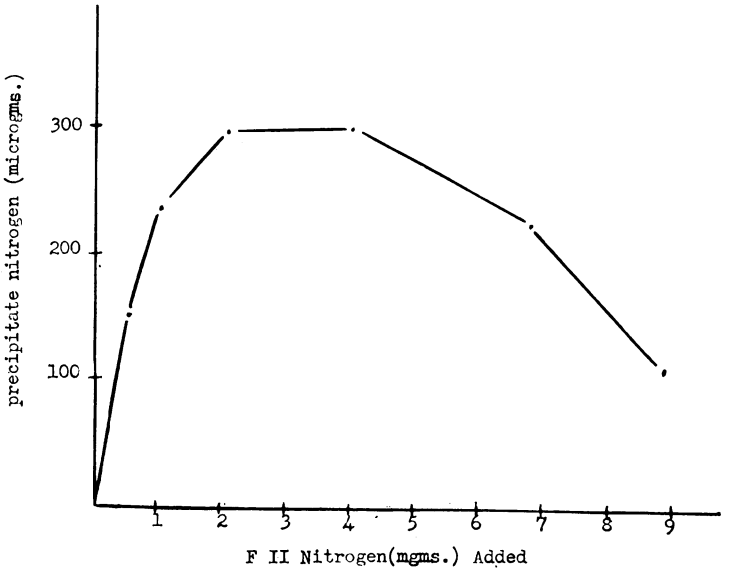

Fig. 2. FII precipitin reaction. Varying amounts of Cohn FII added to constant aliquots of a rheumatoid serum.

trates analytical ultracentrifuge patterns of three rheumatoid sera. The two sera with FII-tanned sheep cell agglutination titers in excess of 1:56,000 demonstrate components which sediment more rapidly than the 19S component. (Serum W.H. with a titer of 1:28,000 is not different from normal human serum in sedimentation studies, demonstrating the relative insensitivity of the ultracentrifuge in the detection of the rheumatoid factor.) The high molecular weight component of very high titered sera (with an S constant of approximately 22), when isolated, possessed rheumatoid factor properties. Furthermore, this heavy component could be made to dissociate into 19S gamma globulin and low molecular gamma globulin with an S constant of 7. With such dissociation, it was the former (19S) which had serological activity. It appears that the rheumatoid factor is a 19S gamma globulin which usually, if not always, exists in whole sera as a soluble complex (22S) formed by reaction with low molecular weight gamma globulin. At the present time, there is no method, other than serologic testing, to differentiate the rheumatoid factor chemically or immunochemically from any other type of 19S gamma globulin in human sera. (A number of bacterial antibodies and iso-agglutinins are known to exist in this 19S fraction.)

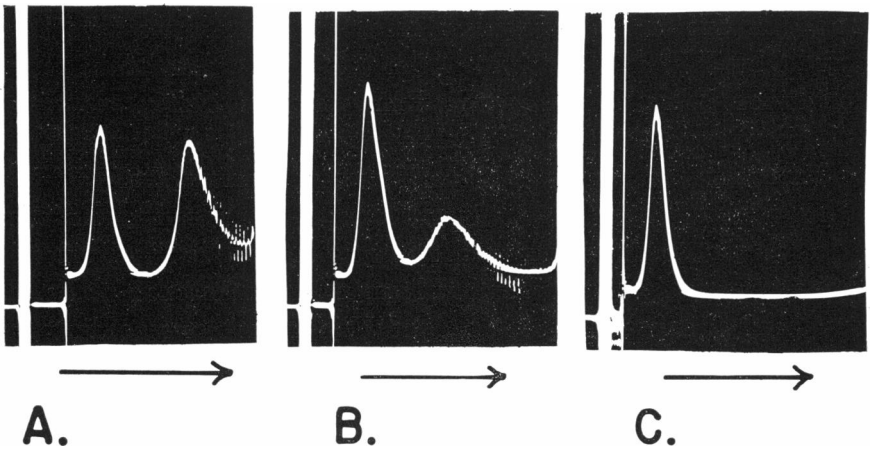

Fig. 3. Analytical centrifuge patterns of three fractions of heated Cohn FII. A and B demonstrate, in addition to 7S gamma globulin, aggregates which sediment rapidly. C contains only 7S gamma globulin.

If one considers our knowledge of the gamma globulin "reactant"—the material with which the rheumatoid factor reacts in serological tests—much of our knowledge derives from the FII precipitin reaction described by Epstein, Johnson and Ragan.[12] As illustrated in Figure 2, the precipitation which results from adding preparations of soluble Cohn FII to rheumatoid sera bears some resemblance to immune precipitin reactions. The precipitate yield compared with the amount of Cohn FII nitrogen added, however, is very small. In brief, the reactive part of Cohn FII in this reaction consists of aggregated gamma globulin. Figure 3 illustrates analytical ultracentrifuge patterns of three preparations of Cohn FII, two demonstrating the presence of components which sediment more rapidly than 7S gamma globulin and the third consisting only of 7S material. Aggregation and subsequent reactivity with the rheumatoid factor can be induced by heating or chemically treating preparations of human gamma globulin which are free of aggregates.[11, 13, 14] Aggregated gamma globulin, in addition to rheumatoid factor reactivity, inactivates serum complement in a manner which qualitatively resembles immune decomplementation,[15-17] induces inflammatory responses in skin which resemble allergic inflammation[18, 19] and precipitates with a newly characterized component of serum complement.[20-22]

There are many important features of this subject which cannot be included in this short presentation. Good general reviews have been presented by Ziff[23] and Vaughan.[24] There are clarifications of the varied reactivity of rheumatoid sera with gamma globulins of different animal species[25] and evidence that different rheumatoid sera display selective reactivity with various genetic types of human gamma globulin.[26, 27]

In consideration of clinical correlations of the rheumatoid factor, the association of the rheumatoid factor with "classical" rheumatoid arthritis (symmetrical small joint involvement, subcutaneous nodules and x-ray changes) approaches 100 per cent. With most test procedures, 70 to 80 per cent of all patients with the clinical diagnosis of rheumatoid arthritis demonstrate positive reactions, but the majority of patients with juvenile rheumatoid arthritis have negative reactions (inhibition tests, which are technically difficult, yield a higher percentage of positive reactions but these procedures have, for the most part, remained research tools). There is evidence, from family and epidemiological studies, which suggests that seropositivity may antedate the clinical onset of rheumatoid arthritis.[28, 29] The so-called variants of rheumatoid arthritis (Marie-Strümpell spondylitis, psoriatic arthropathy, arthritis associated with ulcerative colitis or regional enteritis, agammaglobulinemia arthritis) are, in general, seronegative. Of some potential significance is the growing number of nonrheumatic illnesses significantly associated with the rheumatoid factor. Among this list are some known infectious diseases such as infectious hepatitis,[30] leprosy,[31] syphilis[32] and tuberculosis.[33] Events resulting from chronic infections might have some bearing on the development of the rheumatoid factor.

Recent experimental observations relate the development of a rheumatoid factor-like substance in rabbits to immunization with killed microorganisms over a several-week period.[34] The experimentally induced serum factor which shares many of the properties of the human rheumatoid factor is reactive serologically only with gamma globulin that has been either physically aggregated or gamma globulin contained in an immune complex. Speculation, at the present time, would relate the development of the experimental factor to an immune response against the host's own gamma globulin which has been denatured or altered in the course of *in vivo* formation of immune complexes.

In conclusion, the rheumatoid factor has been isolated and characterized, but its significance relative to the disease, rheumatoid arthritis,

is unknown. The practical value of the serological reactions is limited. They serve primarily as confirmatory tests and infrequently change one's clinical impression or proposed management of an individual case. The hope remains that an understanding of the genesis of the rheumatoid factor may give us some insight into the pathogenesis of rheumatoid arthritis.

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